МОЛЕКУЛЯРНАЯ БИОЛОГИЯ, 2024, том 58, № 1, с. 126–129

МОЛЕКУЛЯРНАЯ БИОЛОГИЯ КЛЕТКИ

УДК 616-006.484:57.023,615.277.3

ВЗАИМОДЕЙСТВИЕ SENP6 С PINK1 СПОСОБСТВУЕТ РЕЗИСТЕНТНОСТИ КЛЕТОК НЕЙРОГЛИОМЫ К ТЕМОЗОЛОМИДУ ЧЕРЕЗ ИНДУКЦИЮ МИТОФАГИИ[#]

© 2024 г. Y. W. Wang[a, §], K. G. Jia[b, §], H. J. Xing[c, §], Y. Pan[d], C. S. Zeng[c], L. Chen[c], Q. J. Su[c], W. T. Shen[e], J. Chen[f], C. Chen[c, *], Q. Cao[g, **], Y. Y. Wang[b, *]**

[a] *School of Medicine of University of Electronic Science and Technology of China, Chengdu, 611730 People's Republic of China*

[b] *Department of Thoracic Surgery, Sichuan Provincial People's Hospital, University of Electronic Science and Technology of China, Chengdu, 610031 People's Republic of China*

[c] *Department of Neurology, The Second Affiliated Hospital of Hainan Medical University, Haikou, Hainan Province, 570100 People's Republic of China*

[d] *Department of Pharmacy, The Second Affiliated Hospital of Hainan Medical University, Haikou, Hainan Province, 570100 People's Republic of China*

[e] *Department of Oncology, Zhujiang Hospital, Southern Medical University, Guangzhou, Guangdong Province, 510280 People's Republic of China*

[f] *Department of Neurology, Danzhou People's Hospital, Danzhou, Hainan Province, 571700 People's Republic of China*

[g] *Department of Assisted Reproductive Medicine, Sichuan Provincial Academy of Medical Sciences and Sichuan Provincial People's Hospital, Chengdu, 611730 People's Republic of China*

[*] *e-mail: 315chencong@163.com*

[**] *e-mail: caoqi19920824@gmail.com*

[***] *e-mail: Wangyouyu121@163.com*

Поступила в редакцию 05.03.2023 г.
После доработки 09.05.2023 г.
Принята к публикации 19.05.2023 г.

Резистентность к темозоломиду считается основной причиной рецидивов и плохого прогноза у пациентов с нейроглиомой. В последнее время появляется все больше данных, свидетельствующих о том, что митофагия вовлечена в развитие лекарственной устойчивости различных типов опухолей. Однако роль и молекулярные механизмы митофагии в резистентности к темозоломиду при глиоме остаются неясными. В проведенном исследовании мы оценили уровни митофагии в резистентных и чувствительных к темозоломиду клеточных линиях. Механизмы, лежащие в основе регуляции митофагии, исследовали с использованием технологии секвенирования РНК. Также проанализирована роль дифференциально экспрессируемых генов при митофагии и резистентности к темозоломиду. Обнаружено, что митофагия вовлечена в развитие резистентности клеток глиомы к темозоломиду. В этом процессе участвует в частности SUMO-специфичная пептидаза-6 (SUMO specific peptidase 6, SENP6), которая индуцирует митофагию. Взаимодействие между SENP6 и главным белком митофагии – PTEN-индуцированной киназой-1 (PINK1) – приводит к снижению степени (SUMO2)илирования PINK1, тем самым усиливая митофагию. На основании полученных результатов можно сделать вывод, что, индуцируя митофагию, взаимодействие SENP6 с PINK1 способствует резистентности клеток глиобластомы к темозоломиду. Таким образом, таргетинг SENP6 или прямое регулирование митофагии можно рассматривать как потенциальные терапевтические мишени для "отмены" резистентности глиомы к темозоломиду.

Ключевые слова: глиобластома, темозоломид, лекарственная устойчивость, SENP6, митофагия, PINK1

DOI: 10.31857/S0026898424010112**, EDN:** NZGQBP

[#]Полный текст статьи на английском языке размещен на сайте издательства Springer – https://link.springer.com/journal/11008

[§]Эти авторы внесли равный вклад.

СПИСОК ЛИТЕРАТУРЫ

1. Lopes M.B.S. (2017) The 2017 World Health Organization classification of tumors of the pituitary gland: a summary. *Acta Neuropathol.* **134**, 521–535.
2. Stupp R., Mason W.P., van den Bent M.J., Weller M., Fisher B., Taphoorn M.J., Belanger K., Brandes A.A., Marosi C., Bogdahn U., Curschmann J., Janzer R.C., Ludwin S.K., Gorlia T., Allgeier A., Lacombe D., Cairncross J.G., Eisenhauer E., Mirimanoff R.O.; European Organisation for Research and Treatment of Cancer Brain Tumor and Radiotherapy Groups; National Cancer Institute of Canada Clinical Trials Group. (2005) Radiotherapy plus concomitant and adjuvant temozolomide for glioblastoma. *N. Engl. J. Med.* **352**, 987–996.
3. Thomas A., Tanaka M., Trepel J., Reinhold W.C., Rajapakse V.N., Pommier Y. (2017) Temozolomide in the era of precision medicine. *Cancer Res.* **77**, 823–826.
4. Zhang J., Stevens M.F., Bradshaw T.D. (2012) Temozolomide: mechanisms of action, repair and resistance. *Curr. Mol. Pharmacol.* **5**, 102–114.
5. Han B., Meng X., Wu P., Li Z., Li S., Zhang Y., Zha C., Ye Q., Jiang C., Cai J., Jiang T. (2020) ATRX/EZH2 complex epigenetically regulates FADD/PARP1 axis, contributing to TMZ resistance in glioma. *Theranostics.* **10**, 3351–3365.
6. Lu C., Wei Y., Wang X., Zhang Z., Yin J., Li W., Chen L., Lyu X., Shi Z., Yan W., You Y. (2020) DNA-methylation-mediated activating of lncRNA SNHG12 promotes temozolomide resistance in glioblastoma. *Mol. Cancer.* **19**, 28.
7. Meng X., Zhao Y., Han B., Zha C., Zhang Y., Li Z., Wu P., Qi T., Jiang C., Liu Y., Cai J. (2020) Dual functionalized brain-targeting nanoinhibitors restrain temozolomide-resistant glioma via attenuating EGFR and MET signaling pathways. *Nat. Commun.* **11**, 594.
8. Tomar M., Kumar A., Srivastava C., Shrivastava A. (2021) Elucidating the mechanisms of Temozolomide resistance in gliomas and the strategies to overcome the resistance. *Biochim. Biophys. Acta Rev. Cancer.* **1876**(2), 188616.
9. Lin Z., Niu Y., Wan A., Chen D., Liang H., Chen X., Sun L., Zhan S., Chen L., Cheng C., Zhang X., Bu X., He W., Wan G. (2021) RNA m^6A methylation regulates sorafenib resistance in liver cancer through FOXO3-mediated autophagy. *EMBO J.* **39**, e103181.
10. Shen W., Zhang W., Ye W., Wang H., Zhang Q., Shen J., Hong Q., Li X., Wen G., Wei T., Zhang J. (2020) SR9009 induces a REV-ERB dependent anti-small-cell lung cancer effect through inhibition of autophagy. *Theranostics.* **10**, 4466–4480.
11. Parzych K.R., Klionsky D.J. (2014) An overview of autophagy: morphology, mechanism, and regulation. *Antioxid. Redox Signal.* **20**, 460–473.
12. Kim K.H., Lee M.S. (2014) Autophagy – a key player in cellular and body metabolism. *Nat. Rev. Endocrinol.* **10**, 322–337.
13. Onorati A.V., Dyczynski M., Ojha R., Amaravadi R.K. (2018) Targeting autophagy in cancer. *Cancer.* **124**, 3307–3318.
14. D'Arcy M.S. (2019) Cell death: a review of the major forms of apoptosis, necrosis and autophagy. *Cell. Biol. Int.* **43**, 582–592.
15. Scrivo A., Bourdenx M., Pampliega O., Cuervo A.M. (2018) Selective autophagy as a potential therapeutic target for neurodegenerative disorders. *Lancet Neurol.* **17**, 802–815.
16. Li W., He P., Huang Y., Li Y.F., Lu J., Li M., Kurihara H., Luo Z., Meng T., Onishi M., Ma C., Jiang L., Hu Y., Gong Q., Zhu D., Xu Y., Liu R., Liu L., Yi C., Zhu Y., Ma N., Okamoto K., Xie Z., Liu J., He R.R., Feng D. (2021) Selective autophagy of intracellular organelles: recent research advances. *Theranostics.* **11**, 222–256.
17. Bernardini J.P., Lazarou M., Dewson G. (2017) Parkin and mitophagy in cancer. *Oncogene.* **36**, 1315–1327.
18. Xie Y., Liu J., Kang R., Tang D. (2020) Mitophagy receptors in tumor biology. *Front. Cell Dev. Biol.* **8**, 594203.
19. Chuang K.C., Chang C.R., Chang S.H., Huang S.W., Chuang S.M., Li Z.Y., Wang S.T., Kao J.K., Chen Y.J., Shieh J.J. (2020) Imiquimod-induced ROS production disrupts the balance of mitochondrial dynamics and increases mitophagy in skin cancer cells. *J. Dermatol. Sci.* **98**, 152–162.
20. Ashrafi G., Schwarz T.L. (2013) The pathways of mitophagy for quality control and clearance of mitochondria. *Cell Death Differ.* **20**, 31–42.
21. Eiyama A., Okamoto K. (2015) PINK1/Parkin-mediated mitophagy in mammalian cells. *Curr. Opin. Cell. Biol.* **33**, 95–101.
22. Heo J.M., Ordureau A., Paulo J.A., Rinehart J., Harper J.W. (2015) The PINK1-PARKIN mitochondrial ubiquitylation pathway drives a program of OPTN/NDP52 recruitment and TBK1 activation to promote mitophagy. *Mol. Cell.* **60**, 7–20.
23. Bravo-San Pedro J.M., Kroemer G., Galluzzi L. (2017) Autophagy and mitophagy in cardiovascular disease. *Circ. Res.* **120**, 1812–1824.
24. Clark E.H., Vázquez de la Torre A., Hoshikawa T., Briston T. (2021) Targeting mitophagy in Parkinson's disease. *J. Biol. Chem.* **296**, 100209.
25. Wei R., Cao J., Yao S. (2018) Matrine promotes liver cancer cell apoptosis by inhibiting mitophagy and PINK1/Parkin pathways. *Cell Stress Chaperones.* **23**, 1295–1309.
26. Liu B., Zhou J., Wang C., Chi Y., Wei Q., Fu Z., Lian C., Huang Q., Liao C., Yang Z., Zeng H., Xu N., Guo H. (2020) LncRNA SOX2OT promotes temozolomide resistance by elevating SOX2 expression via ALKBH5-mediated epigenetic regulation in glioblastoma. *Cell Death Dis.* **11**, 384.
27. Wang Q., Zeng F., Sun Y., Qiu Q., Zhang J., Huang W., Huang J., Huang X., Guo L. (2018) Etk interaction with PFKFB4 modulates chemoresistance

of small-cell lung cancer by regulating autophagy. *Clin. Cancer Res.* **24**, 950–962.

28. Zeng F., Wang Q., Wang S., Liang S., Huang W., Guo Y., Peng J., Li M., Zhu W., Guo L. (2020) Linc00173 promotes chemoresistance and progression of small cell lung cancer by sponging miR-218 to regulate Etk expression. *Oncogene*. **39**, 293–307.
29. Zhang L., Wang L., Hu X.B., Hou M., Xiao Y., Xiang J.W., Xie J., Chen Z.G., Yang T.H., Nie Q., Fu J.L., Wang Y., Zheng S.Y., Liu Y.F., Gan Y.W., Gao Q., Bai Y.Y., Wang J.M., Qi R.L., Zou M., Ke Q., Zhu X.F., Gong L., Liu Y., Li D.W. (2022) MYPT1/PP1-mediated EZH2 dephosphorylation at S21 promotes epithelial-mesenchymal transition in fibrosis through control of multiple families of genes. *Adv. Sci.* (Weinh). **9**, e2105539.
30. Shen W., Zhang X., Fu X., Fan J., Luan J., Cao Z., Yang P., Xu Z., Ju D. (2017) A novel and promising therapeutic approach for NSCLC: recombinant human arginase alone or combined with autophagy inhibitor. *Cell Death Dis.* **8**, e2720.
31. Iwashita H., Torii S., Nagahora N., Ishiyama M., Shioji K., Sasamoto K., Shimizu S., Okuma K. (2017) Live cell imaging of mitochondrial autophagy with a novel fluorescent small molecule. *ACS Chem. Biol.* **12**, 2546–2551.
32. Shihan M.H., Novo S.G., Marchand S.J., Wang Y., Duncan M.K. (2021) A simple method for quantitating confocal fluorescent images. *Biochem. Biophys. Rep*. **25**, 100916.
33. Jensen E.C. (2013) Quantitative analysis of histological staining and fluorescence using ImageJ. *Anat. Rec.* (Hoboken). **296**(3), 378–381.
34. Moore A.S., Holzbaur E.L.F. (2019) Imaging the dynamics of mitophagy in live cells. *Methods Mol. Biol.* **1880**, 601–610.
35. Montgomery M.K., Kim S.H., Dovas A., Zhao H.T., Goldberg A.R., Xu W., Yagielski A.J., Cambareri M.K., Patel K.B., Mela A., Humala N., Thibodeaux D.N., Shaik M.A., Ma Y., Grinband J., Chow D.S., Schevon C., Canoll P., Hillman E.M.C. (2020) Glioma-induced alterations in neuronal activity and neurovascular coupling during disease progression. *Cell Rep*. **31**(2), 107500.
36. Venkataramani V., Tanev D.I., Strahle C., Studier-Fischer A., Fankhauser L., Kessler T., Körber C., Kardorff M., Ratliff M., Xie R., Horstmann H., Messer M., Paik S.P., Knabbe J., Sahm F., Kurz F.T., Acikgöz A.A., Herrmannsdörfer F., Agarwal A., Bergles D.E., Chalmers A., Miletic H., Turcan S., Mawrin C., Hänggi D., Liu H.K., Wick W., Winkler F., Kuner T. (2019) Glutamatergic synaptic input to glioma cells drives brain tumour progression. *Nature*. **573**(7775), 532–538.
37. Venkatesh H.S., Morishita W., Geraghty A.C. (2019) Electrical and synaptic integration of glioma into neural circuits. *Nature*. **573**(7775), 539–545.
38. Jin J. (2019) Interplay between ubiquitylation and SUMOylation: empowered by phase separation. *J. Biol. Chem*. **294**, 15235–15236.
39. Liu C., Zhao H., Xiao S., Han T., Chen Y., Wang T., Ma Y., Gao H., Xie Z., Du L.L., Li J., Li G., Li W. (2020) Slx5p-Slx8p promotes accurate chromosome segregation by mediating the degradation of synaptonemal complex components during meiosis. *Adv. Sci.* (Weinh). **7**, 1900739.
40. Lazarou M., Sliter D.A., Kane L.A., Sarraf S.A., Wang C., Burman J.L., Sideris D.P., Fogel A.I., Youle R.J. (2015) The ubiquitin kinase PINK1 recruits autophagy receptors to induce mitophagy. *Nature*. **524**, 309–314.
41. Geisler S., Holmström K.M., Skujat D., Fiesel F.C., Rothfuss O.C., Kahle P.J., Springer W. (2010) PINK1/Parkin-mediated mitophagy is dependent on VDAC1 and p62/SQSTM1. *Nat. Cell Biol.* **12**, 119–131.
42. Chang H.M., Yeh E.T.H. (2020) SUMO: from bench to bedside. *Physiol. Rev*. **100**, 1599–1619.
43. Hsu C.Y., Yeh L.T., Fu S.H., Chien M.W., Liu Y.W., Miaw S.C., Chang D.M., Sytwu H.K. (2018) SUMO-defective c-Maf preferentially transactivates Il21 to exacerbate autoimmune diabetes. *J. Clin. Invest.* **128**, 3779–3793.
44. Seeler J.S., Dejean A. (2017) 1SUMO and the robustness of cancer. *Nat. Rev. Cancer*. **17**, 184–197.
45. Qiu J., Sheedlo M.J., Yu K., Tan Y., Nakayasu E.S., Das C., Liu X., Luo Z.Q. (2016) Ubiquitination independent of E1 and E2 enzymes by bacterial effectors. *Nature*. **533**, 120–124.
46. Fu H., Liu N., Dong Q., Ma C., Yang J., Xiong J., Zhang Z., Qi X., Huang C., Zhu B. (2019) SENP6-mediated M18BP1 deSUMOylation regulates CENP-A centromeric localization. *Cell Res.* **29**, 254–257.
47. Qian J., Luo Y., Gu X., Wang X. (2013) Inhibition of SENP6-induced radiosensitization of human hepatocellular carcinoma cells by blocking radiation-induced NF-κB activation. *Cancer Biother. Radiopharm.* **28**, 196–200.

Interaction of SENP6 with PINK1 Promotes Temozolomide Resistance in Neuroglioma Cells via Inducing the Mitophagy

Y. W. Wang[1, #], K. G. Jia[2, #], H. J. Xing[3, #], Y. Pan[4], C. S. Zeng[3], L. Chen[3], Q. J. Su[3], W. T. Shen[5], J. Chen[6], C. Chen[3, *], Q. Cao[7, **], and Y. Y. Wang[2, *]**

[1] *School of Medicine of University of Electronic Science and Technology of China, Chengdu, 611730 People's Republic of China*

[2] *Department of Thoracic Surgery, Sichuan Provincial People's Hospital, University of Electronic Science and Technology of China, Chengdu, 610031 People's Republic of China*

[3] *Department of Neurology, The Second Affiliated Hospital of Hainan Medical University, Haikou, Hainan Province, 570100 People's Republic of China*

[4] *Department of Pharmacy, The Second Affiliated Hospital of Hainan Medical University, Haikou, Hainan Province, 570100 People's Republic of China*

[5] *Department of Oncology, Zhujiang Hospital, Southern Medical University, Guangzhou, Guangdong Province, 510280 People's Republic of China*

[6] *Department of Neurology, Danzhou People's Hospital, Danzhou, Hainan Province, 571700 People's Republic of China*

[7] *Department of Assisted Reproductive Medicine, Sichuan Provincial Academy of Medical Sciences and Sichuan Provincial People's Hospital, Chengdu, 611730 People's Republic of China*

**e-mail: 315chencong@163.com*

***e-mail: caoqi19920824@gmail.com*

****e-mail: Wangyouyu121@163.com*

Temozolomide resistance is a major cause of recurrence and poor prognosis in neuroglioma. Recently, growing evidence has suggested that mitophagy is involved in drug resistance in various tumor types. However, the role and molecular mechanisms of mitophagy in temozolomide resistance in glioma remain unclear. In this study, mitophagy levels in temozolomide-resistant and -sensitive cell lines were evaluated. The mechanisms underlying the regulation of mitophagy were explored through RNA sequencing, and the roles of differentially expressed genes in mitophagy and temozolomide resistance were investigated. We found that mitophagy promotes temozolomide resistance in glioma. Specifically, small ubiquitin-like modifier specific protease 6 (SENP6) promoted temozolomide resistance in glioma by inducing mitophagy. Protein-protein interactions between SENP6 and the mitophagy executive protein PTEN-induced kinase 1 (PINK1) resulted in a reduction in small ubiquitin-like modifier 2 (SUMO2)ylation of PINK1, thereby enhancing mitophagy. Our study demonstrates that by inducing mitophagy, the interaction of SENP6 with PINK1 promotes temozolomide resistance in glioblastoma. Therefore, targeting SENP6 or directly regulating mitophagy could be a potential and novel therapeutic targets for reversing temozolomide resistance in glioma.

Keywords: glioblastoma, temozolomide resistance, SENP6, mitophagy, PINK1